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Amanita coacta (*Amanitaceae*, *Agaricales*) with a key to *Amanita* species occurring in Brazil

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Abstract — *Amanita coacta* was described from Amazonas State, Northern Brazil, in 1978 based on a single collection, and since then no more records have been reported. Sixteen collections of this species were made from Parque Estadual das Fontes do Ipiranga, São Paulo State, Southeast Brazil. These collections form a basis for a re-description including range variation of spore size and shape. An artificial dichotomous key to all *Amanita* species occurring in Brazil is also presented.

Key words — *Basidiomycota*, Atlantic forest, taxonomy

Introduction

Amanita Pers. is a well-known genus of *Basidiomycota* with global distribution. It has morphological, anatomical, and developmental characteristics useful for its macroscopical recognition and support inside the family *Amanitaceae* R. Heim ex Pouzar (Drehmel et al. 1999, Oda et al. 1999, Zhang et al. 2004). The genus is characterized by the mycorrhizal habit, hemiangiocarpic development, lamellae that are usually white and free, pallid basidiospores, bilateral hymenophoral trama, and longitudinally acrophysalidic stipe tissue (Bas 1969).

The genus comprises ca. 500 species (Kirk et al. 2001) with 19 taxa mentioned for Brazil, including *A. muscaria* (L.) Lam. (Homrich 1965, Figueiredo et al. 1996, Fosco-Mucci & Yokomizo 1985, Giachini et al. 2000, 2004, Meijer 2001, 2006, Sobestiansky, 2005), *A. pantherina* var. *multisquamosa* (Peck) Jenkins (Giachini et al. 2000, 2004) and *A. rubescens* Pers. (Sobestiansky 2005), all of which were frequently collected in reforestation area with plantations of exotic trees and so likely introduced with seedlings.

Rick (1906) was the first author to describe species of *Amanita* from Brazil. Since then, other publications (Rick 1930, 1937, 1961, Singer 1953, Bas 1978, Grandi et al. 1984, Capelari & Maziero 1988, Bas & Meijer 1993, Pegler 1997, Wartchow et al. 2007, Wartchow & Maia 2007) have contributed to our knowledge of the genus in Brazil.

The study by Bas (1978), although restricted to species collected by Rolf Singer in Amazonas State in Northern Brazil, undoubtedly provides the best knowledge about *Amanita* in Brazil. In his work, eight species with Brazilian types were described, including *A. coacta,* based on a single collection from Amazon Forest.

In this article, *A. coacta* is reported for the first time outside the Amazon region, in a remnant of Atlantic forest of Southeast Brazil. This record represents the second collection of this species since its description. We provide an updated description of the species and present for the first time photos of *A. coacta* showing different morphological patterns. An artificial dichotomous key for *Amanita* species in Brazil is also presented.

Material and methods

The specimens studied were collected at the Parque Estadual das Fontes do Ipiranga, a Atlantic forest remnant in São Paulo State, Southeast Brazil, and deposited at herbarium SP. Fresh specimens were photographed and macro-morphological data were recorded. Color terms are according to Küppers (1979).

For microscopic analyses, the dried material was rehydrated in 70% ethanol followed by 5% KOH or Melzer's reagent. All microscopic illustrations were made with the aid of a drawing tube. The spores were measured in lateral view. The method for metric values follows Tulloss (1993). At the beginning of a set of spores data, the notation "[*a*/*b*/*c*]", where *a*, *b*, and *c*, are integers, is to be read "*a* spores were measured from *b* basidiomata taken from *c* collections". When ranges are provided in spore data in the form "(*m*–)*n*–*o* (–*p*)", where *m*, *n*, *o* and *p* are integers, the values given are to be understood as follows: *m* is the lowest values observed or calculated and *p* the highest. In the range of values observed or calculated, the 5th percentile is *n* and the 95th percentile is *o.* A summary of definitions of biometric variables follows:

W_{cs} = breadth of central stratum of lamella.

W_{st}-near = distance from one side of central stratum to nearest base of basidium.

W_{st}-far = distance from one side of central stratum to the most distant base of basidium on the same side of the central stratum.

L, (**W**) = the range of average lengths (widths) of spores of each basidioma examined.

L', (**W'**) = the average of all lengths (widths) of spores measured.

Q = the ratio of length to width of a spore or the range of such ratios for all spores measured.

Q = the average of Q computed for all basidiomata examined.

Q' = the average of all Q values computed for all spores measured.

DNA sequences of the large subunit (LSU) of nuclear ribosomal DNA were obtained from two collections, for future phylogenetic molecular studies. GenBank accession numbers are cited below the species name at the beginning of the formal description. The dichotomous key presented is based on literature. Generic and infrageneric names and concepts follow Corner & Bas (1962) and Bas (1969).

Results

Taxonomy

Amanita coacta Bas — Figs. 1–2

GenBank FJ236806, FJ236807

Macrocharacters — Pileus 45–69 mm diam, plane-convex, sometimes slightly depressed at centre or concave, dark grayish brown ($N_{80}A_{30}M_{50}$ to $N_{80}A_{60}M_{60}$) at center, somewhat slightly paler toward the margin, margin rather densely sulcate-striate, dry to subviscid, with gray to brownish gray patches of the universal veil scattered or concentrated at center, occasionally lacking volval remnants on pileus. Lamellae free, white, crowded, with or without dark grayish edge; lamellulae scarce or absent, truncate. Stipe 62–100 × 4–8(apex)–8–11(base) mm, subcylindrical, thicker toward the base but without bulb, central, hollow, pale cream with small grayish to grayish-brown fibrils, exannulate. volva at base of stipe, felted-submembranous, with whitish to grayish short fibrils, thin and fragile, easily breakable into grayish patches clearly separated, often forming incomplete transverse zones.

Microcharacters — Basidiospores [340/17/16] 8.7–10(–11.2) × (6.2–)7.5–8.7(–10) µm [**L** = 8.05–10.04; **L'** = 9.46; **W** = 6.85–8.4; **W'** = 7.76; Q = (1.12–)1.15–1.33(–1.4); **Q** = 1.17–1.3; **Q'** = 1.22], subglobose to broadly ellipsoid, rarely ellipsoid, inamyloid, colorless, hyaline, smooth, thin–walled, with large guttule or frequently with precipitated internal content; apiculus lateral to sublateral. Basidia (23–)31–46(–56) × (8.7–)10–12.5(–13.7) µm, clavate, thin-walled, frequently with precipitated internal content, 4-spored, with sterigmata up to 6.2 µm. Pleurocystidia and cheilocystidia absent. Subhymenium cellular, up to 25 µm width, as 2–4 layers of more or less isodiametric to irregular cells, (8.7–)10–18.7(–21) × (6.2–)8.7–12(–15) µm; W_{st}-near = 15–31, W_{st}-far = (32–)37–44. Lamella trama bilateral, slightly divergent, with W_{cs} = 25–37, composed of thin-walled hyphae, hyaline, septate, sometimes branched and slightly inflated, 3.7–8.7(–15) µm diam., and usually with divergent terminal inflated elements up to 35 µm diam. Pileus context undifferentiated, approximately 162 µm thick, composed of thin-walled hyphae, hyaline, septate, 3.7–8.7(–12.5) µm diam. Stipe context longitudinally acrophysalidic, with undifferentiated hyphae 3.7–6.2 µm diam., acrophysalides thin-walled, 162–225 × 27–34 µm, and sometimes with oleiferous hyphae up to 12.2 µm diam. Pileipellis as cutis up to 210 µm thick, composed of thin-walled hyphae, 2.5–10 µm diam., with brown vacuolar pigment and with few conspicuous subradial elements, with or without an ixocutis layer up to 50 µm thick on top, sometimes with distinctive volval remnants. Universal veil on pileus consisting of undifferentiated hyphae 2.5–6.2 µm diam., thin-walled, slightly yellowish, septate, moderately branched, loosely interwoven, with inflated terminals elements and ovoid, globose or subglobose cells, (25–)

30–60(–69) × (19–)22–51(–65) µm, with light brown vacuolar content, thin-walled. Universal veil on basal part of stipe consisting of undifferentiated hyphae, 2.5–7.5 µm diam., thin-walled, slightly yellowish, septate, moderately branched, loosely interwoven, with inflated terminals elements and ovoid, globose or subglobose cells, (20–)25–38(–50) × (16–)20–34(–44) µm with light brown vacuolar content, thin-walled. Clamps absent in all parts examined.

Habitat and substrate — Solitary or in small groups (two to three basidiomata near) on soil in a remnant of Atlantic forest in São Paulo City.

Specimens examined — **BRAZIL. São Paulo State**: São Paulo, PARQUE ESTADUAL DAS FONTES DO IPIRANGA — 29.I.1960, Fidalgo & Furtado s.n. (SP); 21.I.1987, Pegler 3810 (SP); 20.I.1999, M. Capelari et al. s.n. (SP); 20.II.2002, M. Capelari et al. 4148 (SP); 19.XI.2002, M. Capelari & D.M. Vital 4189 (SP); 02.IV.2003, U.C. Peixoto s.n. (SP); 16.II.2004, M. Capelari et al. 4269 (SP); 15.II.2005, M. Capelari & F.V. Neves 4308, 4309 (SP); 03.III.2005, G.R. Leal s.n. (SP); 07.IV.2005 M. Capelari s.n. (SP); 11.I.2007, F. Karstedt & Menolli Jr. 841 (SP); 14.II.2007, Menolli Jr. et al. 99, 100, 101 (SP); 20.IV.2007, Menolli Jr. et al. 126 (SP).

Comments — *Amanita coacta* is a typical neotropical species of subgenus *Amanita*, section *Vaginatae* (Fr.) Quél. due to its inamyloid basidiospores, a densely sulcate pileus margin, a subcylindrical stipe without a bulbous base, and submembranous-felted volva. However, our southeastern Brazilian collections present some differences when compared to the description in the protologue of the type from Amazonas State, Northern Brazil.

Our collections have subhymenium cells slightly shorter and cells of the universal veil on pileus longer than those described in the protologue [25(–35) mm for subhymenium cells and 20–35 mm for volval remnants cells].

The information provided here improves our knowledge of the micro-characters of *A. coacta* including range in spore size and shape and clarifies some characters, such as the lamella trama, which according to the protologue is "impossible to study in type" (Bas 1978). Other macro-morphological variations were not described in the protologue for *A. coacta* because the type is a single basidioma (collected in 1977 by Singer) that Bas had probably not examined fresh. Therefore, we report new morphological variations such as occurrence in small groups predominantly in summer months, a pileus commonly plane-convex and occasionally without volval remnants, an occasionally dark grayish lamellar edge, a stipe with measurable differences between apex and base, and a volva that often forms incomplete transverse zones.

In one collection of *A. coacta* (M. Capelari et al. 4148), at first sight the stipe surface appears to have concentric annuli one-third upwards from the base (Fig. 1j–l). However, this is a misinterpretation because the "annuli" are actually the result of the stipe surface cleaving, probably due to exposure to the sun in an open field instead of the shaded margin of trails in the forest where all other specimens were collected.

Figure 1: *Amanita coacta* collections. a. F. Karstedt & Menolli Jr. 841. b. M. Capelari & D.M. Vital 4189. c. M. Capelari et al. 4269. d. M. Capelari & F.V. Neves 4309. e. Menolli Jr. et al. 99. f–g. Menolli Jr. et al. 126. h. Menolli Jr. et al. 101. i. M. Capelari & F.V. Neves 4308. j–l. M. Capelari et al. 4148. Scale bar = 1 cm.

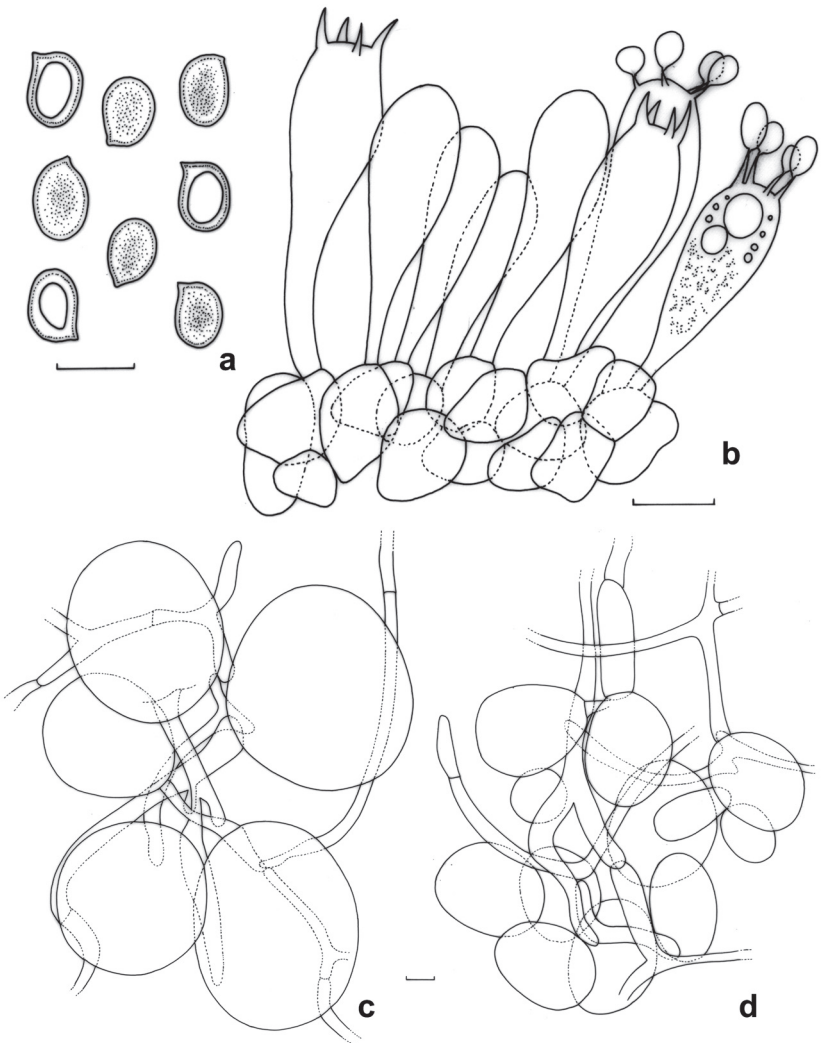

Figure 2: *Amanita coacta* (Menolli Jr. et al. 101). a. Basidiospores (guttulate or with precipitated internal content). b. Basidia and subhymenium. c. Universal veil remnants on pileus. d. Volval elements on stipe. Scale bars = 10 µm.

Such morphological differences can be more easily interpreted when there is a large collection from the same area, and one should not disregard the age of the basidiomata and environmental conditions such as recent rain, humidity

and desiccation. Preliminary molecular nLSU DNA sequence analyses of these collections (data not shown) support the position of *Amanita coacta* in section *Vaginatae* and imply a high similarity among different collections represented by specimens showing a certain degree of morphological variation (e.g., the absence or presence of patches on the pileus).

Amanita coacta is very similar to *A. craseoderma* Bas, also described from Amazonas State, but the latter has (sub)globose basidiospores and pigmented hyphae up to 25 µm wide in the pileipellis (Bas 1978).

The third species described for the same area is *A. crebresulcata* Bas (in section *Amanita*), which differs in having a saccate volva leaving no remnants on pileus. *Amanita crebresulcata* was previously reported for Parque Estadual das Fontes do Ipiranga by Grandi et al. (1984) as "*Amanita* sp. aff. *crebresulcata,*" based on a collection of Fidalgo & Furtado s.n. (SP46749) and by Pegler (1997) also based on SP46749 plus an additional collection, Pegler 3810 (SP214459). Collection SP46749 is poorly preserved, lacking both volva and stipe base, but it retains distinctive pileus patches, a feature that is not characteristic of *A. crebresulcata* and suggesting that the collection more properly represents *A. coacta*. A re-examination of SP214459 likewise demonstrated that it represents *A. coacta* and not *A. crebresulcata*. The records of *A. crebresulcata* for São Paulo State as reported by Grandi et al. (1984) and Pegler (1997) must thus be invalidated.

One other *Amanita* species may occur in this area, represented by a collection containing a single basidioma (07.IV.2005 M. Capelari s.n., SP). This specimen, which microscopically resembles *A. coacta* but differs in its macroscopic appearance, does not appear to have developed completely when collected; therefore, we cannot confirm whether it represents *A. coacta* or a different *Amanita* species.

Species of *Amanita* recorded from Brazil

In the available literature, nineteen taxa of *Amanita* are reported from Brazil. These species are presented in alphabetical order along with their reference of publication and distribution in Brazilian States (Table 1).

When Rick (1906) reported *A. spissa* (Fr.) P. Kumm. from Brazil, he also proposed two varieties for it: *A. spissa* var. *alba* Rick [nom. illegit., non Quél.] and *A. spissa* var. *laeta* Rick. Later Rick (1930, 1937, 1961) cited *A. strobiliformis* (Paulet ex Vittad.) Bertill. and *A. bresadolae* (Rick) Rick [nom. illegit., non Schulzer], all from Rio Grande do Sul State. When reviewing Rick's species, Singer (1953) renamed *A. bresadolae* as *Lepiota crassior* Singer but did not comment on the *A. spissa* varieties published by Rick in 1906. Bas & Meijer (1993) treated *A. spissa* var. *laeta* as a possible synonym of *A. grallipes* Bas & de Meijer described from Paraná State. There is no subsequent mention of *A. spissa*

Table 1: Species of *Amanita* recorded from Brazil, geographic distribution and references list.

Species	Distribution in Brazil[a]	References
Amanita ameghinoi (Speg.) Singer	SP	Pegler (1997)
Amanita campinaranae Bas[b]	AM	Bas (1978)
Amanita chrysoleuca Pegler	PR	Meijer (2006)
Amanita coacta[b]	AM, SP*	Bas (1978), Grandi et al. (1984) as *A. crebresulcata*, Pegler (1997) as *A. crebresulcata*
Amanita craseoderma[b]	AM, RO	Bas (1978), Capelari & Maziero (1988)
Amanita crebresulcata[b]	AM, PR, PE	Bas (1978), Meijer (2006) as "*A.* cf *crebresulcata*", Wartchow & Maia (2007)
Amanita grallipes[b]	PR, RS	Rick (1906) as "*Amanita spissa* var. *laeta*", Bas & Meijer (1993), Meijer (2001, 2006)
Amanita lanivolva Bas[b]	AM	Bas (1978)
Amanita lilloi Singer	PE	Wartchow et al. (2007)
Amanita muscaria	PR, RS, SC, SP	Homrich (1965), Fosco-Mucci & Yokomizo (1985), Figueiredo et al. (1996), Giachini et al. (2000), Meijer (2001, 2006), Giachini et al. (2004), Sobestiansky (2005)
Amanita pantherina var. *multisquamosa*	SC	Giachini et al. (2000, 2004)
Amanita phaea Bas (nom. prov.)[b]	AM	Bas (1978)
Amanita rubescens	RS	Sobestiansky (2005)
Amanita spissa	RS	Rick (1906, 1937, 1961)
Amanita spissa var. *alba*	RS	Rick (1906)
Amanita strobiliformis	RS	Rick (1930, 1937, 1961)
Amanita sulcatissima Bas[b]	AM	Bas (1978)
Amanita xerocybe Bas[b]	AM	Bas (1978)

[a] = Brazilian States: AM = Amazonas, PR = Paraná, PE = Pernambuco, SP = São Paulo, SC = Santa Catarina, RS = Rio Grande do Sul, RO = Rondônia; [b] = type locality in Brazil; * = species recorded in this work

var. *alba* in the literature, and recent authors consider this variety a nomen dubium, since there is neither indication of a specimen in Rick's publication nor preserved material. *Amanita spissa* and *A. strobiliformis* are European taxa that are not included in our key because they were cited only by Rick (1906, 1930, 1937, 1961) and probably do not occur in Brazil.

Amanitopsis plumbea Rick [nom. illegit., non (Schaeff.) J. Schröt.] is another problematic species described by Rick (1937) from Brazil. Bas (1978) studied the lectotype material (J. Rick 12.220, PACA) and considered this taxon as an insufficiently known species, since it was not possible to assign it to either *A. crebresulcata* or *A. coacta;* a third taxon may be involved.

Key to *Amanita* species occurring in Brazil

1. Basidiospores amyloid; pileal margin usually smooth, rarely sulcate-striate; short gills often attenuate ..... (subgenus *Lepidella*) 2
1. Basidiospores inamyloid; pileal margin radially sulcate-striate; short gills nearly always truncate ..... (subgenus *Amanita*) 7

2(1). Pileal margin not appendiculate; surface often deeply colored; basidiospores globose to ellipsoid, mostly < 10 µm, rarely up to 12 µm long; annulus membranous, rarely fugacious ..... (Section *Validae*) 3
2. Pileal margin appendiculate; surface rarely deeply colored; basidiospores globose to bacilliform, rather often > 10 µm; annulus floccose to fugacious ..... (Section *Lepidella*) 5

3(2). Pileus about 6 cm wide, convex to depressed when mature, white to pallid grayish with gray volval crust at center, viscid; basidiospores globose to subglobose, 5.5–7.5 × 5.5–6.5µm ..... *A. campinaranae*
3. Pileus usually wider than 6 cm, conical, hemispheric-convex to plane- convex, rarely depressed 4

4(3). Pileus about 6.5 cm wide, conical, very dark brown with scattered small grayish warts and patches; stipe white above and gray below annulus, with scattered small volval warts; basidiospores characters unknown ..... *A. phaea*
4. Pileus 6–12(–15), convex to applanate or finally depressed, reddish brown or more yellowish and paler, with more or less concentrically arranged whitish to grayish or grayish brown squamules; stipe whitish soon discoloring pinkish and concolor with pileus, with slight volval granulation; basidiospores ellipsoid, 7.5–10 × 4.5–5.5 µm ..... *A. rubescens*

5(2). Pileus hemispherical or conic-convex to plane-convex, uniformly dark brown to somewhat paler grayish brown ..... *A. grallipes*
5. Pileus subglobose or hemispheric to plane-convex white to pale pinkish orange or light beige with patches 6

6(5). Pileus narrower than 4 cm, usually about 2–4 cm; basidiospores subglobose to broadly ellipsoid, mostly < 10 µm, about 7.5–9.5 × 6.5–7.5 µm, basidia 4-spored ..... *A. lilloi*
6. Pileus usually wider than 4 cm; basidiospores ellipsoid, rather often > 10 µm, about 11–13 × 8–10 µm, basidia mostly 2-spored ..... *A. ameghinoi*

7(1). Stipe with a bulbous base; volva usually friable, sometimes limbate; annulus absent or present ..... (Section *Amanita*) 8
7. Stipe without basal bulb; volva saccate to sub-membranous-felted, more rarely friable; annulus absent ..... (Section *Vaginatae*) 13

8(7). Pileus often red, orange or yellow covered with white or yellowish patches or flocculose-pulverulent velar remnants 9
8. Pileus brown, ochraceus brown, brownish, whitish to grayish, covered with gray, white to grayish or brownish ochraceus warts and patches 10

9(8). Pileus 8–18(–25) cm wide, light orange to deeply red, with white patches; basidiospores ellipsoid, 9–11.5 × 6–8 µm ..... *A. muscaria*

9. Pileus 2.5–3.5 cm wide, dry, deep chrome yellow to orange-yellow, covered with yellowish ochraceous, flocculose-pulverulent velar remnants; basidiospores subglobose to ellipsoid, 7–9.5 × 4.5–6 µm ..... *A. chrysoleuca*

10(8). Clamps absent; pileus 4–6.7 cm wide, sordid whitish to ochraceus with brownish ochraceus center; basidiospores globose to subglobose, 8–9 × 7.5–9 µm ..... *A. xerocybe*

10. Clamps present; pileus about 4 cm wide 11

11(10). Annulus fragmentary ..... *A. pantherina* var. *multisquamosa*

11. Annulus always absent 12

12(11). Volva saccate, membranous, gray-brown, enclosing one-quarter to one-third of stipe like a sock ..... *A. lanivolva*

12. Volval remnants at base of stipe arising from upper part of bulb, appressed, pale brownish-gray, with subtomentose-sublanose surface, at one side of stipe forming a thin submembranous limb above bulb ..... *A. sulcatissima*

13(7). Volva friable, forming a dark gray-brown sub-floccose belt at base of stipe and evanescent small dark brown warts on cap ..... *A. craseoderma*

13. Volva saccate or submembranous-felted, white to pale buff or grayish, felted to, at most, appressedly fibrillose on outside limb 14

14(13). Volva membranous, narrowly saccate, white, leaving no remnants on pileus; basidiospores subglobose to broadly ellipsoid, rarely ellipsoid, (8–)8.5–10(–11) × (6.5–)7–8.5(–9)µm ..... *A. crebresulcata*

14. Volva felted sub-membranous, tending to break up into small appressed flat gray patches at stipe base and around cap center; basidiospores subglobose to broadly ellipsoid, 8.7–10(–11.2) × (6.2–)7.5–8.7(–10) µm ..... *A. coacta*

According to the classification of Corner & Bas (1962), the Brazilian species are distributed in the two subgenera *Amanita* and *Lepidella* and in four sections *Amanita*, *Vaginatae*, *Validae,* and *Lepidella*. The infrageneric classification of *A. lanivolva* should be regarded with caution, however. Bas (1978) first treated this species in section *Vaginatae* based on its possession of a saccate volva. Simmons et al. (2002) later emphasized the presence of a small, but distinct basal bulb in collections of Guyana to refer *A. lanivolva* to section *Amanita*, although the presence of a saccate volva is uncommon within the section.

Of the new species described by Bas (1978), only *A. craseoderma* (Capelari & Maziero 1988) and *A. crebresulcata* (Meijer 2006, Wartchow & Maia 2007) have been found and mentioned outside Amazonas State. Simmons et al. (2002) also recorded *A. lanivolva* and *A. xerocybe* from Guyana.

According to Bas (1978), a provisional name was given for *A. phaea*, because basidiospores were lacking in the type material; other characters, such as a smooth and non-appendiculate pileus margin, friable volva, and attenuate lamellulae, are enough to classify it in the section *Validae*.

The results of this paper strongly support the necessity of collecting and studying *Amanita* in South America because very little is known about its neotropical species.

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